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CONTRIBUTIONS FROM THE BERMUDA BIOLOGICAL STATION FOR RESEARCH. — No. 15.

REGENERATION IN THE BRITTLE-STAR OPHIOCOMA PUMILA, WITH REFERENCE TO THE INFLUENCE OF THE NERVOUS SYSTEM.

By Sergius Morgulis.

With a Plate.

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REGENERATION IN THE BRITTLE-STAR OPHIOCOMA PUMILA, WITH REFERENCE TO THE INFLUENCE OF THE NERVOUS SYSTEM.[1]

By Sergius Morgulis.

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In animals with a well differentiated nervous system all functions are so intimately associated with this system that the severing of the connection between an organ and its nervous supply leads to a loss of function, and at times also to an atrophy of the organ itself. Furthermore, the nervous system exercises an important rôle in regulating the interrelation of parts of the organic complex, so that interference with, or loss of, one function may — through the nervous system — lead to a more or less profound disturbance of another function. Indeed cases of abnormalities or monstrosities are not infrequently attributable to some disturbance in the nervous system.

Leaving aside entirely those instances which fall within the scope of embryology, Herbst, it may be recalled, found in the crustacean Porcellana that whether there was regenerated an eye or an antenna in place of an extirpated eye depended wholly upon whether or not the optic ganglion had been injured by the operation. It may also be recalled that the exposure of the cut end of the nerve cord is a condition *sine qua non* for the regeneration of the head in the earthworm, as was discovered by Morgan.

The evidence concerning this problem of the influence of the nervous system is, however, very conflicting in some important points, and so far as vertebrates are concerned there is apparently no agreement among writers, although the opinion is strong that the central nervous

[1] I am under obligation to Dr. E. L. Mark, both for the opportunity of research which I enjoyed at Bermuda, and for the careful revision of the manuscript.

system does not exert any appreciable influence upon the process of regeneration.

While working at the Bermuda Biological Station for Research last summer, I undertook a study of some phases of this problem on the brittle-star Ophiocoma pumila with a view to determining certain points, especially whether or not there exists a relation between the nervous supply and the rate with which a part of an organism regenerates. The brittle-stars present certain advantages for such a study, (1) because the operation is not connected with a profuse bleeding, (2) because there are several similar parts which may be operated upon simultaneously, and (3) because the same animal with its five similar arms can be used

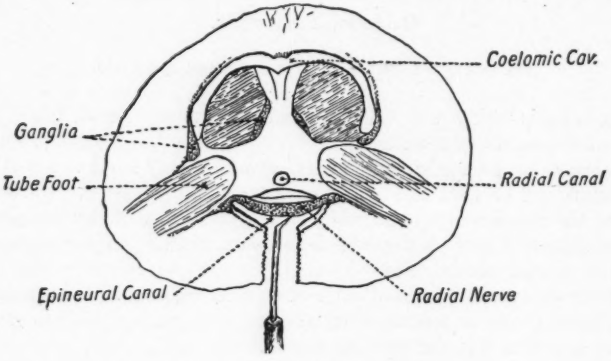

FIGURE *A*.

both for the experiment and for the control, the variations incident to the use of different individuals being thus eliminated.

Unfortunately the want of an abundant material and the great mortality among the operated animals prevented me from obtaining a decisive answer to all the questions which interested me, and the facts to be presented here form merely the beginning of a more extensive investigation which I hope to pursue at the earliest opportunity.

Before discussing my experiments and their outcome I will say a word about the anatomy of the nervous system of the brittle-star and about the method of operation. The central nervous system of the Ophiuroids, unlike that of the star-fish, is a deeply seated organ, and consists of the ring-nerve around the oesophagus, and radial nerves extending out from the ring-nerve into each of the five arms. The ring-nerve and the radial nerves are really double structures, one system being super-

imposed upon the other; they are usually designated as ectoneural and hyponeural systems. There are in addition many ganglia and an elaborate peripheral system of nerves, but we shall not be concerned with the latter.

The operation consisted in destroying a small portion of the radial nerve in order to break its connection with the ring-nerve, and was performed in the following manner: The calcareous plate on the oral surface of an arm was first punctured with a needle (see Figure *A*) a very short distance from the disc. In this way an opening was established leading into the canal in which the nerve lies. If released at this phase of the operation, the animal would crawl away, using all its arms, and behaving in an absolutely normal fashion, showing thus that the injury was not serious. Next, a red hot needle was introduced into the opening already made, burning the nerve at that particular spot, as will be seen from the diagram. To prevent the needle from injuring the deeper portions of the arm, its point was bent at an angle of 45°. After this operation the animal would crawl away, but would use only the uninjured arms, while the injured arm would be practically paralyzed and curled up about the point of injury, being dragged along passively. In nearly all cases when the wound was not made deep the arm was not cast off even at the end of thirty days, when the animals were preserved. Whenever the wound was made too deep, the arms were subsequently cast off.

When this preliminary operation had been accomplished, the arm was cut off at about the middle of its length. In a number of animals another arm with the nerve intact was also cut off at about the middle, this serving as a control for the arm with an injured nerve. In every case the arms with the radial nerve intact regenerated from the cut surface, and so far as I could ascertain, they regenerated quite normally. On the other hand, if the radial nerve was injured before cutting off the arm,[2] the latter in the course of thirty days regenerated only a small stump, which might easily be overlooked unless the specimen were examined closely. Figures 1 to 5 of the Plate represent several brittle-stars in all of which the same results appear. Where the radial nerve was left intact, a long new part was regenerated (Plate, Figures 1, 3, and 5), whereas if the nerve was destroyed near the disc, so little new tissue was formed that it is difficult to recognize it at all. The interesting thing in this connection, however, is that in cases where the animal threw off the arm at the place of injury to the nerve, there was absolutely no regeneration from the cut surface thus produced

[2] The place of injury to the nerve is indicated by a cross in Figures 1 to 5.

(as will be seen from Figure 2), while other arms in the same specimen with the nerve intact have regenerated normally.[3]

There are three possible interpretations of this phenomenon : (1) It may be essential for the regeneration of an arm that the cut nerve should present a free end, as was the case with the earthworm. Or, (2) it may be possible that the undestroyed portion of the radial nerve between the point of injury and the cut end of the arm could furnish sufficient impetus to cause a slight regeneration. Finally, (3) the explanation may be that in those cases where the wound was made deep the continuity of all parts of the nervous tissue present in both the superficial and deeper portions of the arm was destroyed. Which of these explanations is the correct one must be decided by future experiments.

Experiments by injuring the ring-nerve have not yet been successful, owing to the great difficulty of such an operation.

Before concluding I wish to mention some of the observations made on the *rate* of regeneration of arms. This matter was examined from two standpoints : the relation of the rate of regeneration, first, to the level at which the arms were cut, and, secondly, to the number of arms removed. A few specimens represented by Figures 6 to 14 show the nature of the results. If we compare the rate of regeneration of arms cut at the base with that of those cut near the middle of their length, making proper allowance for individual variations, it will be almost impossible to say which regenerates most. On the other hand, comparing arms cut off at the base, or at the middle (Figures 7, 11, 13, and 14), with those cut off near the tip (Figures 6 and 10), the difference in the rates of regeneration becomes very striking. The total amount regenerated during the same period is much greater in the case of the shorter stubs than in the case of the very long one ; indeed, it would not be an exaggeration to say that the greatest regeneration from the arm cut near its tip does not exceed the least regeneration from one cut at its middle. These results are in perfect agreement with Miss King's results on the regeneration of arms in Asterias.

As regards the second point — the relation of the rate of regeneration to the number of removed arms — my experiments with brittle-stars from which 1, 2, 3, 4, or even 5, arms had been removed by being cut off at the base, do not fully conform to Zeleny's rule, which was based

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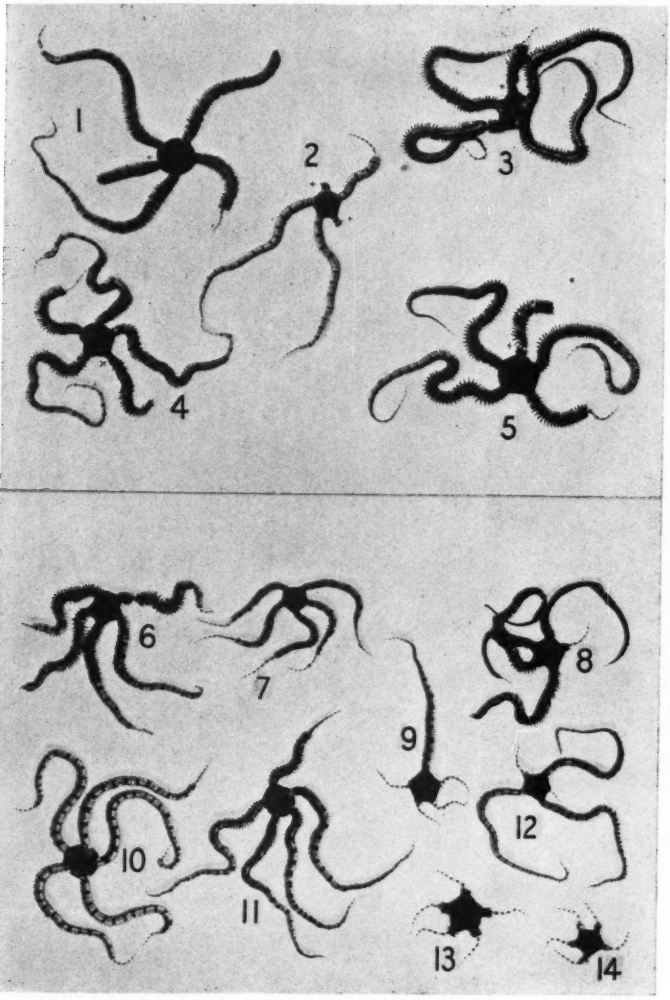
[3] Miss H. D. King, working on the regeneration of the star-fish, found that on cutting the arms horizontally just above the vertebral ridge the edges of the dorsal parts curled under, but did not regenerate, while the ventral parts, containing the radial nerve, reproduced a new dorsal surface.

on his study of regeneration in the brittle-star Ophioglypha lacertosa. He formulated his rule in these words: "The rate of regeneration of a removed arm increases as the number of uninjured arms still remaining decreases." According to my own observation specimens of Ophiocoma pumila with 1 to 3 arms removed regenerate in the course of thirty days new arms ranging in length from 10 to 11 mm., while those deprived of 4 or 5 arms regenerate arms from 10 to 13 mm. long. It will also be observed that there was an equally rapid regeneration in the two animals, one with all five arms cut off at the middle and the other with one arm only thus cut off (Figures 7 and 11).

It is evident from this that there is some correlation between the degree of injury and the rate of regeneration, but that this relation is not of the nature of a close parallelism, such as is suggested by Zeleny's rule. Furthermore, it is stated that in Ophioglypha lacertosa "the regenerated lengths are on the whole at least twice as great in Series IV, where four arms were removed, as in Series I, where only one arm was removed." This, again, differs from my results in Ophiocoma pumila, where the regenerated lengths never presented such wide variations.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